The Avian Influenza Epidemic in Europe in 2003

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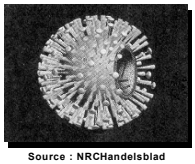
Background on Avian Influenza

- Avian Influenza is caused by influenza A viruses
- Disease first observed in Italy in 1878
- RNA virus with 8 genome fragments
- Classification into subtypes on the basis of two surface proteins:
  - Hemagglutinin (H)
  - Neuraminidase (N)
- In 1955 virus responsible for the disease identified as one of the Influenza A viruses



Source : NRCHandelsblad

Background on Avian Influenza

- Avian Influenza viruses infecting poultry can be subdivided into two groups on the basis of severity of disease after experimental infection
  - Low Pathogenic Avian Influenza (LPAI)
  - Highly Pathogenic Avian Influenza (HPAI) or Fowl Plague (historic name)

03M-0212.4 *Bwa*

Background on Influenza A Viruses

16 H-subtypes and 9 N-subtypes

- Not all combinations appear in nature :
  - in poultry mainly H1, H5, H7 and H9
  - in man mainly H1, H2 and H3
  - in swine mainly H1 or H3 in combination with N1 and N2
  - in horses mainly H3 or H7 in combination with N7 and N8
- HPAI outbreaks in domestic poultry limited to subtypes H5 and H7 and by exception H10 - but not all viruses of these subtypes will cause HPAI !

03M-0212.5 *Bwa*

Background on Avian Influenza

- Wild waterfowl (especially ducks, swans, geese, gulls) carry many different Influenza A viruses - among others LPAI virus of subtype H5 and H7
- First isolation in 1961 in *Sterna hirundo* in South Africa after massive mortality

- From 1972 on it became clear : waterfowl carry huge amounts of AI viruses with large variation in subtypes
- AI viruses are spread with these migrating birds all over the world and are subject to genetic changes (reassortment)

03M-0212.6 *Bwa*

AI in Domestic Poultry World-Wide

- Relatively rare in chickens since 1955 and in many cases with mild clinical symptoms
- Scotland - Italy - USA - Israel - Australia - Hong Kong - USSR - Belgium - France
- In Turkeys from the 1960s on an increasing economical problem Scotland – USA – Canada - UK - Italy

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AI in Domestic Poultry World-Wide

- In domestic ducks known since 1956
- In spite of many isolations of AI viruses in wild ducks : domesticated ducks are ignored as source and reservoir of AI viruses

Canada USA Australia Israel Czechoslovakia Poland Ukraine Yugoslavia Hungary USSR Hong Kong England Italy Belgium Germany

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HPAI Virus Isolates Since 1959

1. Scotland / 59 (H5N1)
2. England / 63 (H7N3)
3. Ontario / 66 (H7N3)
4. Victoria / 66 (H7N7)
5. Germany / 79 (H7N7)
6. England / 79 (H7N7)
7. Pennsylvania / 83 (H5N2)
8. Ireland / 83 (H5N8)
9. Victoria / 85 (H7N7)
10. England / 91 (H5N1)
11. Victoria / 92 (H7N3)
12. Queensland / 94 (H7N3)
13. Mexico / 94 (H5N2)
14. Pakistan / 94 (H7N3)
15. New South-Wales / 97 (H7N4)
16. Hong Kong / 97 (H5N1)
17. Italy / 97 (H5N2)
18. Italy / 99 (H7N1)
19. Hong Kong / 02 (H5N1)
20. Netherlands / 03 (H7N7)

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AI History in the Netherlands

- Fowl Plague first described in 1924 in the Netherlands in same area as in 2003
- Large outbreak in 1927 described
  - Spread between farms over distances of 0.5 to 2 km
  - Indirect contacts via humans suggested
  - Introduction into the Netherlands via Italy suggested
- In former Dutch colony The East Indies one of the oldest AI virus strains isolated
  - A / Fowl Plague virus / Dutch / 1927 / H7N7
  - still used by WHO as reference strain for H7N7

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AI History in the Netherlands

- Several LPAI virus strains isolated in 1980s in broilers: subtype H1N1 - H3N2 - H9N1
- LPAI subtype H7 isolated in parakeets in 1994
- LPAI subtype H5 isolated in emus exported to USA in 1994
- LPAI subtype H5 isolated in ostriches exported to Denmark in 1996

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AI in Wild Waterfowl in the Netherlands

- In 1997 National Influenza Center (prof. Osterhaus - Rotterdam) started collecting faeces samples from wild waterfowl along the most important migration routes from Scandinavia via Western Europe to West-Africa
- Primarily : wild ducks – geese – gulls
- Approximately 5,000 faeces samples per year

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AI in Wild Waterfowl in the Netherlands

- An AI virus subtype H7N3 was isolated from a mallard (*Anas platyrhynchos*) in 2000 in The Netherlands
- An AI virus subtype H11N7 was isolated from a shoveler (*Anas clypeata*) in 1999 in the Netherlands
- The AI virus subtype H7N7 of the 2003 epidemic is probably a fusion of above-mentioned viruses in ducks to a LPAI virus

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Global AI Situation in 2002-2003

- Outbreaks in domestic poultry :
  - USA : LPAI subtype H7N2
  - Chile : HPAI subtype H7N3 and H5N2
  - Italy : LPAI subtype H7N3
- Agreement within European Union on Monitoring in 2002
  - Random sampling of domestic poultry in densely populated areas in member states in 3-year-period
  - Focus on LPAI virus subtype H5 and H7
- Results sampling would be decisive for possible inclusion of LPAI virus subtype H5 and H7 in list-A diseases of O.I.E.

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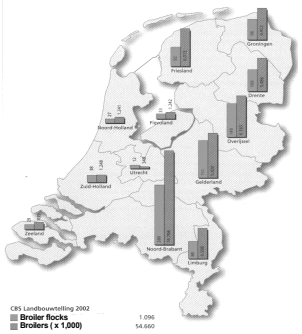
Distribution of Broilers and Broiler Flocks in the Netherlands



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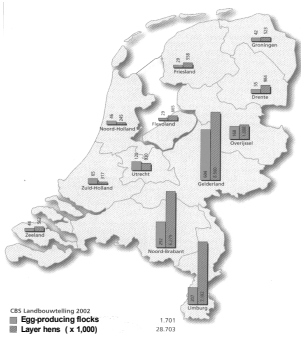
Distribution of Egg-Producing Flocks and Layer Hens in the Netherlands



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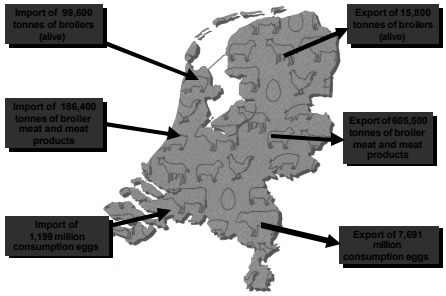

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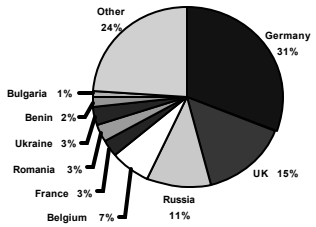
Dutch Export of Poultry Meat (687.000 Tonnes) in 2002



Source: Product Boards for Livestock and Meat 2003

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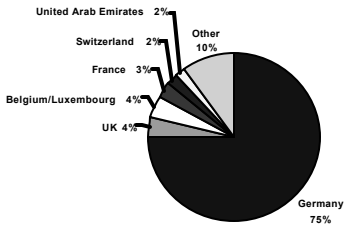
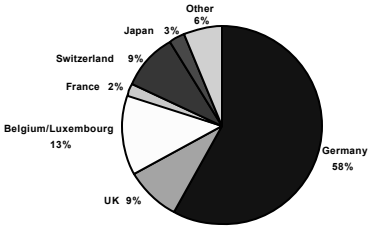
Dutch Export of Consumer Eggs (5,600 Million Eggs) in 2002



Source: Product Boards for Livestock and Meat 2003

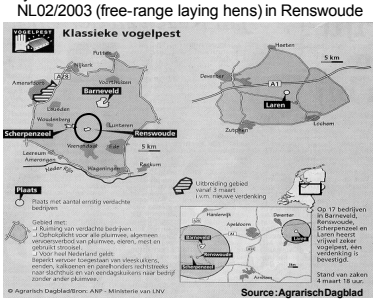
Dutch Export of Egg Products (65,000 Tonnes) in 2002



Source: Product Boards for Livestock and Meat 2003

Detection First HPAI Outbreaks in Netherlands

Saturday 22 Feb : decrease in feed and water intake in outbreak NL02/2003 (free-range laying hens) in Renswoude



Source: Agrarisch Dagblad

Detection First HPAI Outbreaks in Netherlands

- 23 Feb. : continued decrease in feed and water intake in NL02/2003 and start increased mortality (1.5%)
  - Farmer thinks of *E.Coli* infection
  - Contact by phone with Technician of breeding company
- 24 Feb. : no feed and water intake at all and 5% mortality
  - Clinical signs : slight respiratory problems – severe diarrhea – severe apathy – severe drop in egg production
  - Technician visits flock : diagnosis —— Turkey Rhinotracheitis (TRT)
  - Seven dead chickens sent to post-mortem examination to support diagnosis

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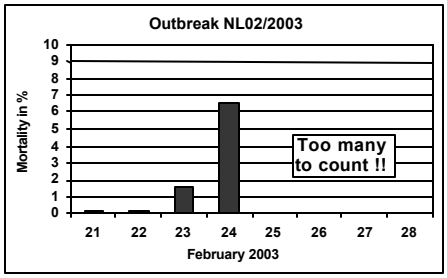
Development of Mortality in NL02/2003



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Detection First HPAI Outbreaks in Netherlands

- 24 Feb. : necropsy results NL02/2003
  - 5 birds with peritonitis & 2 birds with slight tracheitis
  - Preliminary diagnosis : *E.coli* infection
  - Supplementary investigation : Bacteriology and Newcastle Disease (ND) test
- 24 Feb. : start increased mortality in NL05/2003 (flock located across the street (< 250 meter) at NL02/2003) in Renswoude
- 25 Feb. : supplementary investigation NL02/2003
  - *E.coli* : positive ND test : negative
  - On basis necropsy results : breeding company veterinarian (without clinical inspection) prescribes antibiotic (OTC) and brings boxes to farmer to sent other birds to post-mortem

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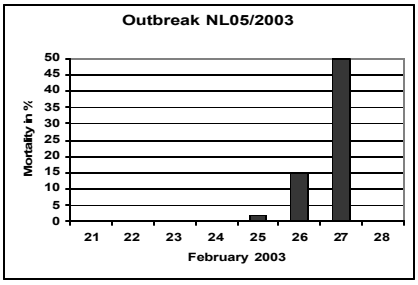
Detection First HPAI Outbreaks In Netherlands

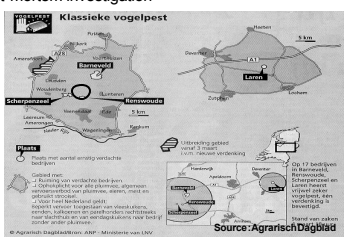
- 25 Feb : increased mortality observed by farmer in outbreak NL05/2003
  - Contact by farmer with Feed-Mill company → sending a veterinarian in afternoon
  - Veterinarian contacts Animal Health Service (AHS) by telephone and describes clinical situation : parent stock with hemorrhagic and red-inflamed trachea
  - AHS confirms by phone preliminary diagnosis of veterinarian : Infectious Laryncho Tracheitis (ILT)
  - AHS recommends veterinarian to send poultry to post-mortem to support a diagnosis
- Unfortunately no birds are sent for post-mortem investigation until Friday 28 February when almost no birds were alive !!

Development of Mortality in NL05/2003



Detection First HPAI Outbreaks In Netherlands

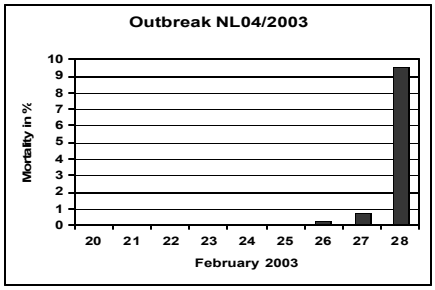
- 26 Feb : dead birds from outbreak NL02/2003 and NL04/2003 (flock located in Scherpenzeel) our brought for post-mortem investigation



Detection First HPAI Outbreaks in Netherlands

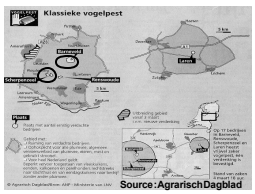
- 26 Feb : dead birds from outbreak NL02/2003 and NL04/2003
  - Clinical signs: mortality – decrease in production – diarrhea
  - Necropsy results: peritonitis – swollen liver and swollen spleen
- On the basis of anamnesis and post-mortem results preliminary diagnosis : *Salmonella gallinarum* infection (partly because in same area earlier two cases of *S. gallinarum* diagnosed with same clinical symptoms)
- Supplementary laboratory investigations started to confirm *S.gallinarum* infection

Development of Mortality in NL04/2003



Detection First HPAI Outbreaks In Netherlands

- 27 Feb : Supplementary laboratory investigations from NL02/2003 and NL04/2003 on *S.gallinarum*: negative
- Dead birds from outbreak NL01/2003 (located in Scherpenzeel) and NL03/2003 (located in Barneveld) are brought for post-mortem investigation



Detection First HPAI Outbreaks in Netherlands

- 27 Feb. : Dead birds from outbreak NL01/2003 and NL03/2003 are brought for post-mortem investigation
  - Clinical signs : acute mortality – decrease in feed/water intake
  - Necropsy results: peritonitis – swollen liver and disturbed ovary
- Mortality in NL05/2003 is dramatic

  As a therapy Flumequine is added to drinking water

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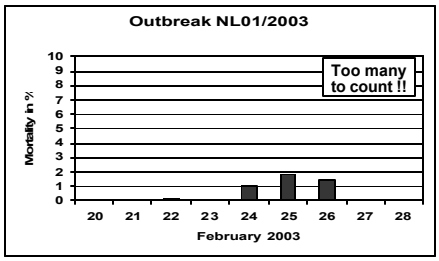
Development of Mortality in NL01/2003



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Detection First HPAI Outbreaks in Netherlands

- 28 Feb. : Supplementary laboratory investigations from NL02/2003 and NL04/2003 on *S.gallinarum* still negative
- In order to exclude other possible causes: tests on ND and AI are initiated on tissue samples from NL01/2003 and NL02/2003
- In addition : veterinary poultry expert from AHS is sent to both poultry flocks for clinical inspection
- Report in afternoon by telephone of dramatic clinical situations by veterinary poultry expert coincides with a positive test result from immunofluoresence test (IFT) for Avian Influenza on tissue samples from NL01/2003 and NL02/2003

ALARM !

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Course of the AI Epidemic in Netherlands

In the weekend of 1-2 March hectic talks at the Emergency Management Staff of the Ministry of Agriculture on eradication of a possible HPAI outbreak

Soon becomes clear that there is not enough depopulation capacity

↓

Partly because a common method – Prussic acid gas – is not allowed anymore since introduction of occupational health regulations

Source : Agrarisch Dagblad

Contact with Belgian and Danish authorities for their experience with CO and CO_2 gassing of poultry units

Course of the AI Epidemic in Netherlands

- Control measures implemented – actions undertaken to prevent spread of AI virus:
  - Nation wide ban on gathering of poultry and other birds
  - Nation wide transport ban of live poultry and hatching eggs
  - Prohibition of export of live poultry and hatching eggs
  - Obligation to keep poultry inside – ban on free-range housing
  - Stringent and nation-wide biosecurity measures on poultry holdings

Source: Agrarisch Dagblad

Course of the AI Epidemic in Netherlands

- Control measures implemented – actions undertaken to prevent spread of AI virus (continued):
  - Establishment of protection - surveillance – and buffer zones around presumed outbreaks
  - Start of clinical inspections in restriction areas from a temporary local crisis center
  - Pre-emptive slaughter of high-risk contact flocks and poultry flocks in one-kilometer radius of outbreak

Source: Agrarisch Dagblad

In Total 5 Different Culling Methods Used

- CO_2 gassing of sealed poultry units

Source : Agrarisch Dagblad

In Total 5 Different Culling Methods Used

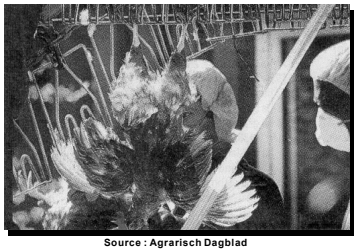
- CO gassing of sealed poultry units (only during day-time and under supervision of Fire-department)

Snel ruimen met grotere vergassingswagens

Source : Agrarisch Dagblad

In Total 5 Different Culling Methods Used

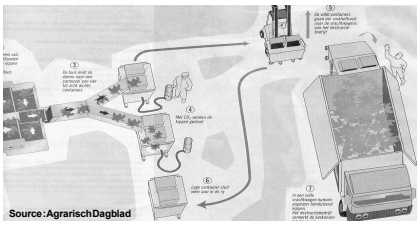
- Mobile slaughter plant



Source : Agrarisch Dagblad

In Total 5 Different Culling Methods Used

- Animal Euthanasia Device (AED) for small number of birds (250 –900 birds per hour)



Source: AgrarischDagblad

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In Total 5 Different Culling Methods Used

- Injection with T61 (medicinal product) for individual birds (pet and hobby birds)

Source: AgrarischDagblad

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Course of the AI Epidemic in Netherlands

- 2 March : CIDC-Lelystad announces results of IVPI (Intra-venous Pathogenicity Index) tests on SPF-birds: HPAI and AI subtype H7
- 3 March : the group of prof. Osterhaus (Erasmus University Rotterdam) announces subtyping of AI virus : AI subtype N7
- 4 March : the first outbreak is depopulated : NL03/2003
- Outbreak NL01/2003 and NL02/2003 officially declared infected

Source :AgrarischDagblad

03M-0212.4i Elbers

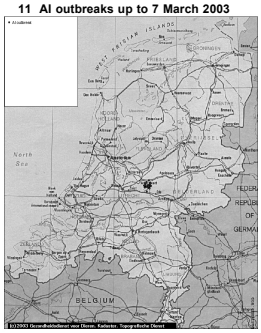
Course of the AI Epidemic in Netherlands

- 5 March : 6 other flocks are officially declared infected and a total of 8 poultry flocks are depopulated
- 6 March : 4 other poultry flocks depopulated

Kippenruimers genieten van korte pauze in lentezon

Source :AgrarischDagblad

11 AI outbreaks up to 7 March 2003

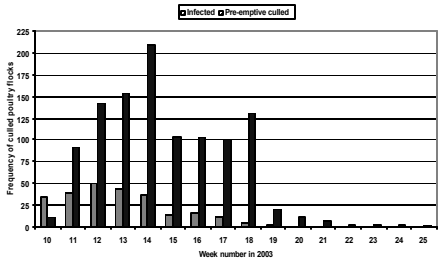


Course of the AI epidemic in Netherlands

- Pre-emptive slaughter was delayed in the first weeks due to depopulation capacity problems
- Capacity problems in processing culled birds (incinerator) maximum capacity 1,400 tonnes per day
- At peak of epidemic : 500,000 - 1 million birds culled per day

Source:AgrarischDagblad

Distribution of Culled Poultry Flocks in Time During the AI Epidemic in the Netherlands In 2003



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What Caused the Epidemic to Take Off ?

- Already a considerable number of flocks were infected before detection of first outbreak creating possibility for transmission to other flocks
- Delay in first weeks of depopulation of infected flocks after first suspicion
- Delay in pre-emptive slaughter of high-risk contacts and flocks in area with 1-kilometer radius around outbreak

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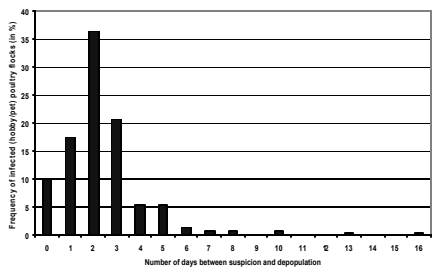
Distribution of Number of Days Between Suspicion and Depopulation of Infected (Hobby/Pet) Poultry Flocks During the AI Epidemic in the Netherlands In 2003



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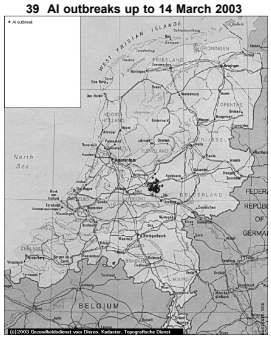
39 AI outbreaks up to 14 March 2003



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72 AI outbreaks up to 21 March 2003

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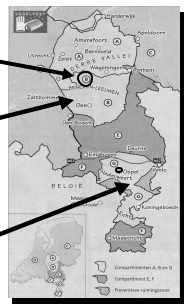
Course of the AI Epidemic

- On 25 March the epidemic escaped in a southern direction out of the "Gelderse Vallei" to Beneden Leeuwen
- As a precaution, priority was given to depopulate complete B-sector between the large rivers to prevent escape to southern provinces
- Unfortunately, the epidemic escaped to the south-eastern part of the Netherlands with a high density of poultry flocks beginning of April



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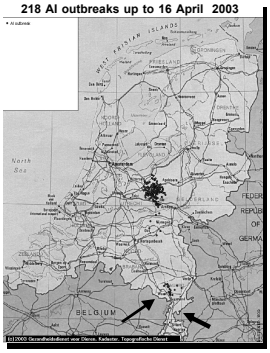
Several poultry flocks (among others several turkey flocks) got infected in the southern Netherlands, sometimes close to the border with Belgium and Germany

218 AI outbreaks up to 16 April 2003



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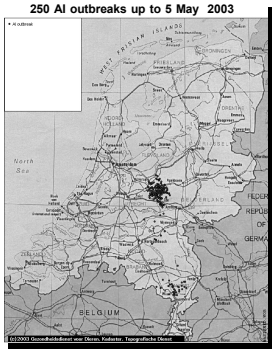
In total, 255 outbreaks were culled with ± 30 million birds

Last outbreak announced on 23 May

Majority of outbreaks concerned layer flocks:

- **22 hobby/pet bird holdings**
- **18 Turkey flocks**
- **2 duck flocks**
- **1 geese flock**
- **3 broiler flocks**

250 AI outbreaks up to 5 May 2003



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Analysis Effectivity Control Measures

R_h = mean number of secondary cases (flocks) per infectious flock during its entire infectious period

$R_h < 1$ requirement for epidemic to fade out

$R_h = \beta * 1 / \alpha$

β : transmission velocity parameter

influenced by number of contacts - hygiene - vaccination

$1 / \alpha$: mean length of infectious period

influenced by timely detection of outbreak after introduction in flock

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Analysis Effectivity Control Measures

- For calculation of R_h the following assumptions were made (supported by experimental transmission studies) :
  - A flock becomes infectious for other flocks two days before observation of first increased mortality;
  - Virus introduction has taken place two days before a flock becomes infectious for other flocks.

Analysis Effectivity Control Measures

Area: "Gelderse Vallei"

Period	R_h	Mean infectious period in days
up to 1 March	5.6	11.8
1 – 8 March	1.03	5.8
8 – 14 March	± 1	5.1

Analysis Effectivity Control Measures

Area: Southern Netherlands

Period	R_h	Mean infectious period in days
up to 10 April	2.0	± 5
10 – 20 April	0.99	± 5

Conclusions Effectiveness Control Measures

- Initial control measures (stand still, transport ban etc.) significantly reduced R_h from 6 to 1
- During the epidemic it was very difficult to decrease R_h much lower than 1
- In order to bring down R_h to ± 0.5 it is necessary to decrease mean infectious period by 50% to approximately 2.5 days

  → **extremely difficult**
- Other option : decrease β by vaccination

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Infection of Swine with AI Virus H7N7

- Initially there was no indication for possibility of swine infection with H7N7 → **Literature**

- After some time during the epidemic reasons for change of opinion:
  - H7N7 grew on swine kidney -cells causing Cytopathogenic Effect
  - Persons with intensive contacts with infected poultry developed conjunctivitis - most certainly caused by infection and replication of AI virus

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Infection of Swine with H7N7 : RISKS

- Re-assortment of H7N7 with other Influenza-strains in swine producing a pathogenic strain for humans (pandemics of 1918 and 1968)
- Circulation within swine causing clinical symptoms and economical damage comparable with Porcine Influenza strains
- Risks for poultry as swine would act as reservoir and re-infection would occur from the reservoir to poultry

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Investigation : Swine Infection with H7N7

- Blood samples collected from swine on 13 high risk herds : infected poultry herds with also swine on premises
- Poultry was declared infected and culled 3-5 weeks before blood sampling of swine
- In 5 herds all swine were sampled (smaller herds)
- In 8 herds from a sample of swine blood collected (60 - 118 samples)
- Haemagglutination inhibition (HI) test
  - Based on experience with HI tests in swine a threshold titer of 1 : 40 was defined as positive

Investigation : Swine Infection with H7N7

Outbreak	# sows	# fatteners	# samples	# positive
4		430	61	16 (26%)
15		306	306	0
16	1	61	62	0
20	69	454	118	6 (5.1%)
21		376	376	0
27		250	250	0
35		400	60	0
36		438	60	5 (8.3%)
39		800	71	0
55		430	60	0
60		260	260	0
66		735	72	4 (5.6%)
83	12	116	60	9 (15%)

Investigation : Swine Infection with H7N7

- Results indicated H7N7 was introduced into swine population
- The 5 likely infected swine herds were re-sampled 11 days after their first sampling in order to measure true transmission within herd
- All individual swine > 4 weeks of age were blood sampled and tested
- Per herd 60 oropharyngeal swabs taken for antigen detection by PCR (for AI viruses in general and H7 specific)

Investigation : Swine Infection with H7N7

Outbreak	% positive 1st sampling	% positive 2nd complete sampling
4	26.0 %	29.0 %
20	5.1 %	2.5 %
36	8.3 %	6.9 %
66	5.6 %	7.7 %
83	15.0 %	14.0 %

03M-0212.ppt Elbers

Investigation : Swine Infection with H7N7

- In outbreak # 20, # 36, and # 66 seropositives were found scattered across the herds – rarely more than one pig per pen
- In outbreak # 4 (29% seroprevalence) seropostives were correlated with feeding of broken eggs from infected poultry in 2 compartments
- All PCR swabs were negative

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Conclusions Swine Infection with H7N7

- Swine in mixed herds with infected poultry are at risk for introduction of AI subtype H7N7
- (Ongoing) transmission between pigs unlikely
- No evidence found that virus is maintained in the swine herds after removal of source of infection (poultry)
- Likely that no residual AI virus was present in any of the swine herds tested

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Chronology of AI Situation – Belgium in 2003

- Since report of 1st AI -outbreak in the Netherlands

Crisis unit put in state of alert in Belgium

- Control measures implemented – actions undertaken to prevent introduction of AI virus from the Netherlands:
  - Nation wide ban on gathering of poultry, other birds
  - Nation wide transport ban of live poultry and hatching eggs
  - Stringent and nation-wide biosecurity measures on poultry holdings
  - Limited access to poultry holdings, especially for any person in contact with poultry in Netherlands

Chronology of AI Situation – Belgium in 2003

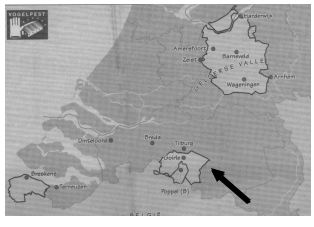
Control measures implemented (continued):

- Increased vigilance with regard to poultry and egg transports at the Dutch-Belgian border
- Notification of any disease or increased mortality for which HPAI can not be excluded
- Prohibition of medical treatment of poultry unless samples have been sent to regional Animal Health Service
- Tracing on of all Belgian high-risk contact holdings (import of live birds or hatching eggs / indirect contact with infected Dutch premises)
- Establishment of protection and/or surveillance zones as a result of outbreaks in the Netherlands neat Belgian border

Chronology of AI Situation – Belgium in 2003

- First suspicion of AI outbreak on 11 March in 12,000 broiler flock in Ravels near Poppel (< 0.5 km from Dutch Border)

2,000 birds died on one day !



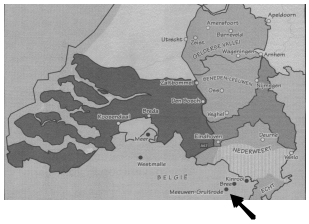
Source: Agrarisch Dagblad

Chronology of AI Situation – Belgium In 2003

- Poultry flock in Ravels depopulated within 24 hours of suspicion
- Backyard poultry in an area with one-kilometer radius of suspect flock destroyed
- On 24 March definite diagnostic test result for suspicion in Ravels : negative

Chronology of AI Situation – Belgium in 2003

- Second clinical suspicion of AI outbreak on 15 April in 10,500 parent poultry stock in Meeuwen-Gruitrode (< 14 km from Dutch border)



Source: Agrarisch Dagblad

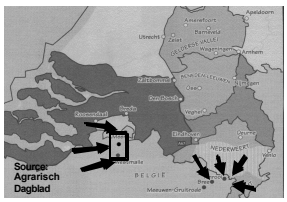
Chronology of AI Situation – Belgium in 2003

Lab confirmation on 18 April : positive HPAI subtype H7

Source: Agrarisch Dagblad

Chronology of AI Situation – Belgium in 2003

- Within a period of 2 weeks a total of 8 HPAI outbreaks
  - Four more outbreaks in area around first outbreak
  - Three other outbreaks 60 km west of primary outbreak area



Chronology of AI Situation – Belgium in 2003

- In the period of 16 April to 15 May 2003
  - A total of 129 poultry flocks depopulated
  - A total of 3.2 million birds destroyed and disposed

Source: Agrarisch Dagblad

Chronology of AI Situation – Germany in 2003

Suspicion of AI outbreak on 9 April in 32,000 broiler flock in Viersen (± 16 km from Dutch Border)

Chronology of AI Situation – Germany in 2003

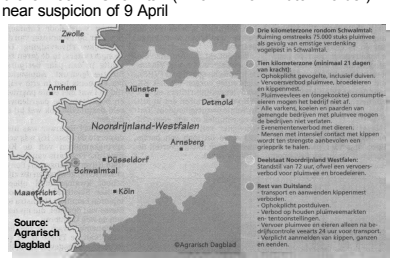
- Exclusion zone set up in a half-mile radius around suspected flock

Source: Agrarisch Dagblad

- Few days later laboratory confirmation: negative

Chronology of AI Situation – Germany in 2003

Suspicion of AI outbreak in evening on 8 May in 30,000 broiler flock in Swalmtal (± 10 km from Dutch Border) near suspicion of 9 April



Source: Agrarisch Dagblad

Chronology of AI Situation – Germany in 2003

Flock was culled next day as were contact flocks (total 80,000 birds)

Source: Agrarisch Dagblad

Chronology of AI Situation – Germany in 2003

- Within radius of 3 km around outbreak all poultry flocks pre-emptively culled within 48 hours
- A protection zone with a radius of 10 km around outbreak, and a surveillance zone with a further radius of 10 km
- Laboratory confirmation on 13 May: HPAI subtype H7
- No further cases reported since 13 May and restriction measures lifted as of 24 June 2003

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Source: AgrarischDagblad

03M-0212.86 Elbers